

EpiPlant 2019

Plant epigenetics meeting,
February 6th - 8th, 2019

Hosted by
Institut de biologie moléculaire des plantes (IBMP) – CNRS,
Strasbourg (France)

6
8 February 2019

EpiPlant 2019

PLANT EPIGENETICS MEETING
STRASBOURG | FRANCE



The Scientific organizing committee

Fredy Barneche, Moussa Benhamed, Nicolas Bouché, Martin Crespi, Laurence Drouard, Jean Molinier

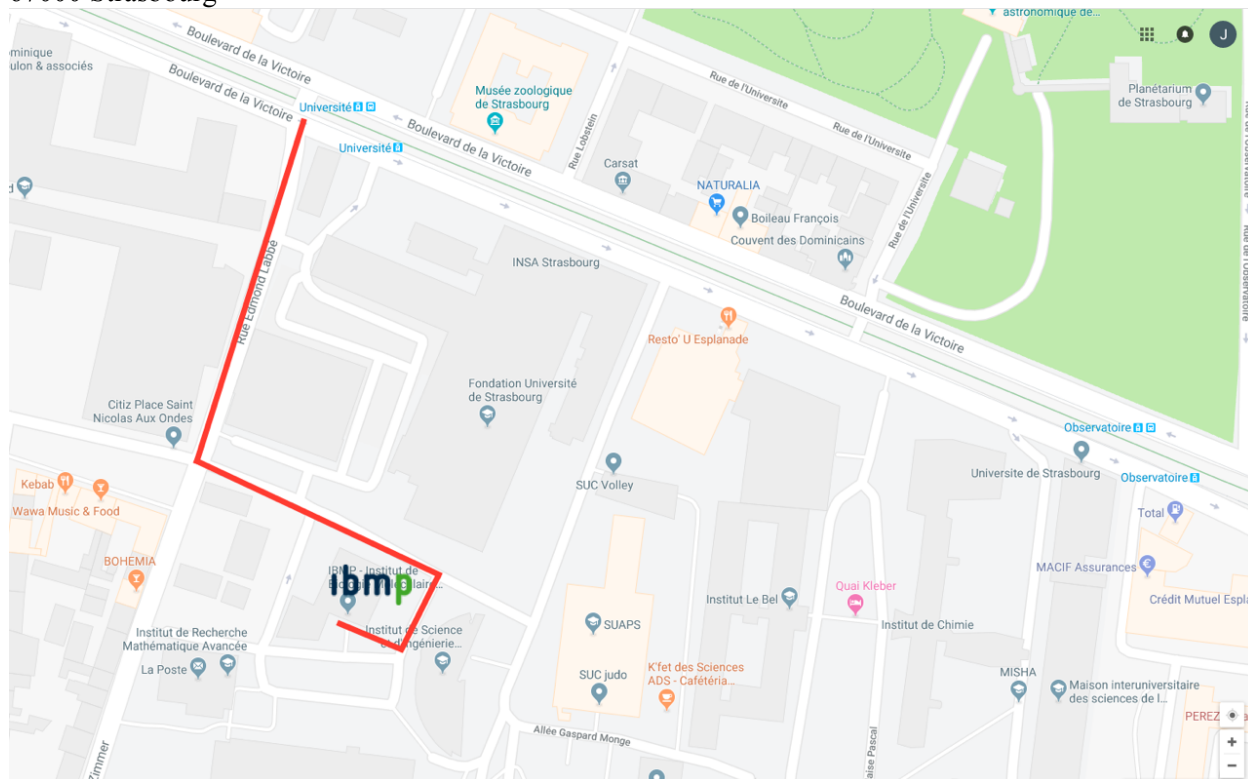
Local organizing committee

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The Symposium will be held at IBMP in Strasbourg, France.

LOCATION

Institut de biologie moléculaire des plantes
CNRS - IBMP
12 rue du Général Zimmer
67000 Strasbourg



Adapted from Google Maps

HOW TO GET TO IBMP BY TRAM

Tram C (Direction Neuhof Rodolphe Reuss) or Tram F (Direction Place d'Islande)
Stop "Université"

HOW TO GET TO STRASBOURG - BY AIR

Strasbourg-Entzheim International Airport

Route de Strasbourg

67960 ENTZHEIM

Tel 03 88 64 67 67 - Fax 03 88 68 82 12

E-mail : information@strasbourg.aeroport.fr - Internet : www.strasbourg.aeroport.fr

◆ Taxis

Taxis are available at the airport until 11 p.m. (the arrival time of the last flight).

◆ Train shuttles

The airport is connected to Strasbourg railway station by a train shuttle which runs up to 4 times per hour, with a journey time of 9 minutes.

For info: Tel. 0800 77 98 67 (freephone within France) - www.ter-sncf.com/alsace

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- Basel-Mulhouse airport (Distance: 130 km from Strasbourg)

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Wednesday February 6th

12.30-14.00

Registration

14.00

Opening

Laurence Drouard (*IBMP, Strasbourg*)

14.15

Kick off GDR EPIPLANT

Jean Molinier (*IBMP, Strasbourg*)

15.00

Keynote lecture

Thomas Sexton (*IGBMC, Illkirch*)

3D genome folding and transcriptional regulation

16.00 Refreshment

16.30

Epigenetic regulations in response to the environment

Chair: Wen-hui Shen (*IBMP, Strasbourg*)

François Parcy (*LPCV, Grenoble*)

The LEAFY floral regulator, a pioneer transcription factor ?

Iva Mozgová (*IPMB, Biology Centre CAS*)

Novel functions of PRC2 during embryo-to-seedling transition in *Arabidopsis*

Clara Richet-Bourbousse (*ENS, Paris*)

Modeling the *Arabidopsis* DNA damage transcriptional response to identify new players in chromatin-based DNA repair mechanisms

Stéphane Maury (*LBLGC, Orléans*)

EPIgenetic variation and plasticity in forest TREES (EPITREE)

Fredy Barneche (*ENS, Paris*)

Light signaling drives multilevel chromatin state changes during *Arabidopsis* development

Johan Zicola (*MPI, Cologne*)

Targeted DNA methylation represses two enhancers of *FLOWERING LOCUS T* in *Arabidopsis thaliana*

Clémentine Vitte (*GQE, Le Moulon*)

Genome-wide hypermethylation of TEs and centromeres following low temperature exposure in maize

19.00 Welcome Cocktail

Thursday February 7th

08.30

Functions of non-coding RNAs

Chair: Martin Crespi (*IPS2, Gif*)

Hervé Vaucheret (*IJPB, Versailles*)

Distinct components of the RdDM pathway contribute to sense transgene PTGS initiation and PTGS-induced DNA methylation in *Arabidopsis*

Quentin Thomas (*Plant and Environmental Sciences - Copenhagen*)

Plant transcriptome heterogeneity revealed by Transcript Isoform Sequencing (TIF-Seq)

Pascal Gamas (*LIPM, Toulouse*)

Dynamics of DNA methylation during symbiotic nodule development in *Medicago truncatula*

Thomas Roulé (*IPS2, Gif*)

Ecotype-related long non-coding RNAs in epigenetic regulation of root growth

10.00-10.30 Coffee break

David Baulcombe (*University of Cambridge*)

The tomato epigenome

Guillaume Hummel (*IBMP, Strasbourg*)

Differential epigenetic regulation of clustered nuclear tRNA genes expression in *Arabidopsis thaliana*

Julie Leclercq (*AGAP, Montpellier*)

Post-transcriptional and transcriptional regulations by small RNAs in *Hevea* latex cells

Hélène Proust (*IPS2, Gif*)

Impact of DCL3 and RTL1 RNases in small RNA dynamics and epigenetic regulations in the nitrogen-fixing symbiosis of *Medicago truncatula*

12.30 Lunch

14.15

Chromatin Dynamics

Chair: Moussa Benhamed (*IPS2, Gif*)

Stefan Grob (*University of Zurich*)

Transgene Silencing in 3D - How a Chromosomal KNOT Can Inactivate Foreign DNA Elements

Christophe Laloi (*BIAM, Aix-Marseille*)

Topoisomerase VI participates in an insulator-like function that prevents heterochromatin spreading into euchromatin islands

Aline Probst (*GREC, Clermont-Ferrand*)

Histone H3.3 deposition in *A. thaliana*

Lorenzo Concia (*IPS2, Gif*)

Chromosome architecture of hexaploid wheat (*Triticum aestivum* L.)

Diagenode

16.00-16.30 Coffee break

Frédéric Pontvianne (*LGDP, Perpignan*)

Ribosomal RNA genes distribution and expression organize nucleolus associated chromatin domains

Christel Carles (*LPCV, Grenoble*)

Regulation of chromatin mark changes for organogenesis in *Arabidopsis*

Taline Elmayan (*IJPB, Versailles*)

A transgene locus mimics transposon *de novo* silencing

Laura Ferrafiat (*IBMP, Strasbourg*)

The NRPD1 N-terminus contains a Pol IV-specific motif that is critical for genome surveillance in *Arabidopsis*

18.30 Poster viewing + Discussions + Snack Bretzel Drinks

Friday February 8th

08.30

Transgenerational epigenetic variation

Chair: Nicolas Bouché (*IJPB, Versailles*)

Mélody Nicolau (*LGDP, Perpignan*)

Role of Plant Mobile Domain proteins in regulation of gene expression

Pierre Bourguet (*GRED, Clermont-Ferrand*)

Pol epsilon links DNA replication with gene silencing, heterochromatin structure and DNA methylation

Mélanie Jubault (*IGEPP, Rennes*)

Arabidopsis natural and induced epialleles associated with quantitative resistance to clubroot

Vincent Colot (*ENS, Paris*)

Chromatin-guided transposition promotes the creation of epigenetically-sensitive mutations in genes controlling adaptive traits

10.00-10.30 Coffee break

Hua Jiang (*IPK, Gatersleben*)

AT-hook proteins increase histone modification H3K9me2 independently of DNA methylation

Diep T. N. Tran (*ENS, Paris*)

A novel RdDM-based reporter system to dissect transgenerational epigenetic inheritance in *Arabidopsis*

Bart Rymen (*IBMP, Strasbourg*)

Epigenetic reprogramming associated with wound-induced organ regeneration

Daniel Zilberman (*JIC, Norwich*)

Stable epigenetic inheritance of DNA methylation through pathway integration

12.00

Concluding remarks

Take away sandwiches

Abstracts of the main talks

Thomas Sexton	p. 8
François Parcy	P 9
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David Baulcombe	p. 12
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Vincent Colot	p. 15
Daniel Zilberman	p. 16

3D genome folding and transcriptional regulation

Thomas Sexton

Institute of Genetics and Molecular and Cellular Biology (IGBMC), Illkirch, France

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Eukaryotic genomes are highly compacted in order to be contained within the nucleus, but in a highly organised manner that enables access of transcription, replication and repair machinery to the appropriate genes according to developmental context. For a long time, microscopic studies have correlated transcriptional activity with gene position relative to other nuclear landmarks, but the advent of molecular biology methods coupled to high-throughput sequencing (the “Hi-C” and related technologies) has greatly refined our understanding of finer-scale chromosome folding. This talk will focus on two scales of chromatin topology which are found to be exquisitely correlated with gene expression control: loops that physically juxtapose gene promoters with long-range regulatory elements, and autonomously folded regions (“topologically associated domains”; TADs) that appear to delimit local chromatin state and activity. However, despite intensive studies, causal (if any) and mechanistic links between chromatin topology and transcriptional control remain unclear. Using mouse thymocyte development and embryonic stem cells as model systems, we have uncovered complex developmental dynamics of chromatin loops and (to a lesser extent) TADs, and present evidence for these dynamics, in some cases, to be responsible for transcriptional control.

The LEAFY floral regulator, a pioneer transcription factor ?

François Parcy¹, Arnaud Stigliani, Renaud Dumas, Camille Sayou, Emmanuel Thevenon, Jérémy Lucas, Eugenia Brun-Hernandez, Jeanne Loue-Manifel, Chloe Zubieta

¹Physiologie Cellulaire et Végétale – Grenoble, CNRS : UMR5168, Commissariat à l'Énergie Atomique et aux Énergies Alternatives (CEA) - Grenoble, Institut national de la recherche agronomique (INRA), , University of Grenoble Alpes (UGA)

The development of flowers requires the expression of whole sets of genes that are maintained silent before the floral transition. In *Arabidopsis*, LEAFY is a master transcription factor responsible for initiating such gene expression programs. Molecular genetics combined with structural biology and genomics showed that the SAM (Sterile Alpha Motif) oligomerization domain of LEAFY allows the binding to closed chromatin regions, suggesting LEAFY might act as a pioneer factor. We will present to latest result of our laboratory to test this attractive hypothesis.

Light signaling drives multilevel chromatin state changes during *Arabidopsis* development

Fredy Barneche

IBENS, Ecole Normale Supérieure, CNRS UMR 8197 Paris, France

Light not only constitutes an essential energy source for photosynthesis but also provides spatio-temporal information used by plants to trigger adaptive responses. In particular, the first exposure to light of a germinating plantlet induces chloroplast biogenesis and phototrophic growth. Within the few hours, this photomorphogenic transition involves reprogramming of thousands of genes' expression in cotyledon cells, many of them undergoing pioneering rounds of transcription. This event is paralleled by dynamic changes of histone mark profiles over regulated genes, of heterochromatin organization and of nucleus size. In previous studies, we also reported a ~3-fold increase of global RNA Polymerase II activity, suggesting that a switch from relatively quiescent to more active transcriptional status is induced by light in most cotyledon nuclei. We aim at assessing the functional consequences of such higher-order changes and the molecular mechanisms driving them. We will present how the chromatin-associated photomorphogenic regulator DET1 influences both heterochromatin dynamics and histone H2B monoubiquitination homeostasis by controlling expression of a linker histone variant and the stability of a SAGA-like deubiquitination module, respectively. Hence, we describe a signaling path that modulates the epigenome landscape, hypothetically contributing to adjust chromatin status to the cell transcriptional activity.

Distinct components of the RdDM pathway contribute to sense transgene PTGS initiation and PTGS-induced DNA methylation in *Arabidopsis*

Hervé Vaucheret

Institut Jean-Pierre Bourgin, INRA, AgroParisTech, CNRS, Université Paris-Saclay, 78000, Versailles, France

During sense-transgene post-transcriptional gene silencing (S-PTGS), DNA methylation is established in the transgene transcribed portion, but its role remains unclear. Here, we show that the upstream components of the RNA-directed DNA methylation (RdDM) pathway, CLSY1, NRPD1, RDR2 and DCL3, are not required for S-PTGS-induced DNA methylation, whereas the downstream RdDM components NRPE1, DRD1 and DRM2, as well as the core S-PTGS component RDR6, are required, suggesting that RDR6-dependent siRNAs trigger S-PTGS-induced DNA methylation in an NRPE1-, DRD1- and DRM2-dependent manner, similar to de novo DNA methylation of reactivated transposons. Nevertheless, none of these RdDM components are required for induction of spontaneous S-PTGS, or for silenced tissue to transmit a systemic S-PTGS signal, suggesting that DNA methylation is a consequence, not a cause, of S-PTGS. However, NRPD1 and RDR2 are required for induction of systemic S-PTGS upon grafting of nonsilenced scions onto silenced rootstocks. NRPD1 and RDR2 are also required for spontaneous S-PTGS induced by a *vcs* mutation that allows the accumulation of aberrant RNAs. We propose that NRPD1 and RDR2 stimulate the production of transgene aberrant RNAs, thus promoting spontaneous S-PTGS in a *vcs* background, as well as induction of systemic S-PTGS in nonsilenced scions grafted onto silenced rootstocks.

The tomato epigenome

David Baulcombe

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It is an oversimplification to think of epigenetics in plant and animal genomes as simply defence against the mutagenic effects of transposable elements. Epigenetics does silence these mobile DNAs and it does prevent their damaging influence on genes and chromosomes but there are also more diverse effects - both positive and negative - on the transposon and the host genome.

To characterise the influence of an epigenome on a plant genome we are using tomato as a model system. Our approaches involve gene-edited mutants and genetic knock down in epigenetic pathways involving RNA-directed DNA methylation (*NRPD1*, *NRPE1*) and other functions (*DDM1*, *CMT3*, *KYP*) and wide cross hybrids of cultivated and wild tomato.

These approaches lead to the conclusion that the epigenome is dynamic with features being gained and lost over time. These features may influence gene expression and, in some extreme instances, they are associated with non-Mendelian inheritance patterns that resemble paramutation in maize and other species.

Transgene Silencing in 3D – How a Chromosomal *KNOT* Can Inactivate Foreign DNA Elements

Stefan Grob and Ueli Grossniklaus

Institute of Plant and Microbial Biology, University of Zurich, Switzerland

Cells require elaborate mechanisms to efficiently pack chromosomes in the nucleus, while still allowing access to the genetic information. In addition, three-dimensional (3D) chromosome architecture is linked to epigenetic processes and transcriptional activity. Despite progress in the field, well-established cases of functional relationships between transcription and 3D chromatin architecture remain rare. We previously identified a 3D chromatin structure in *Arabidopsis* termed the *KNOT*, in which ten genomic regions (*KEEs*) physically contact each other.

Here we show that *KEEs* are involved in the silencing of transgenes. Transgenes integrated in the genome can fold towards the *KNOT*, coinciding with their transcriptional silencing. Thus, transgene integration can lead to significant perturbation of 3D chromosome architecture. Regions adjacent to the insertion sites are not subjected to silencing, despite their dislocation within the nucleus. This novel silencing mechanism, termed *KNOT*-linked Silencing (KLS) may act independently of previously described silencing mechanisms, as we cannot observe any significant contribution of small RNAs and DNA methylation. KLS is heritable across generation and shows *trans*-silencing effects, as the introduction of *KNOT*-silenced transgenes can lead to the silencing of previously active transgenes.

Ribosomal RNA genes distribution and expression organize nucleolus associated chromatin domains

Picart-Piccolo A.^{1,2}, Picault N.^{1,2}, Picart C.^{1,2}, Pontvianne F^{1,2}

¹ CNRS, Laboratoire Génome et Développement des Plantes, UMR 5096, 58 Avenue P. Alduy, 66860 -Perpignan, France

² Univ. de Perpignan Via Domitia, Laboratoire Génome et Développement des Plantes, UMR 5096, 58 Avenue P. Alduy, 66860 - Perpignan, France

Genome interactions can occur beyond chromosome territories and organize around nuclear bodies. Recent studies revealed how the nucleolus organizes high order chromatin structure of certain chromosome regions displaying heterochromatic features. We are currently studying the mechanisms involved in the heterochromatin organization and their impact on gene expression using *Arabidopsis thaliana* as model organism. We isolated nucleoli using fluorescence-activated cell sorting (FACS) and identified Nucleolus-Associated chromatin Domains (NADs) by deep sequencing. In wild-type, NADs are primarily genomic regions with heterochromatic signatures and include transposable elements (TEs), sub-telomeric regions, and mostly inactive protein-coding genes. However, NADs also include active ribosomal RNA (rRNA) genes and the entire short arm of chromosome 4 adjacent to them (Pontvianne *et al*, 2016). Analyses of NADs in mutant affected in rRNA genes expression or in lines with different amount of rRNA genes reveal how affecting rRNA genes nuclear positioning and expression affect NADs identity. In the light of our data and additional studies made in animal cells, we will discuss the potential role of the ribosomal RNA chromosome location in the establishment of inactive chromatin association with the nucleolus.

Pontvianne F, Carpentier MC, Durut N, Pavlišťová V, Jaške K, Schořová S, Parrinello H, Rohmer M, Pikaard CS, Fojtová M, Fajkus J and Sáez-Vásquez J. (2016) Identification of nucleolus-associated chromatin domains reveals the role of the nucleolus in the 3D organisation of the *A. thaliana* genome. *Cell Reports*. August 9 (16) doi:10.1016/j.celrep.2016.07.016

Chromatin-guided transposition promotes the creation of epigenetically-sensitive mutations in genes controlling adaptive traits

Leandro Quadrana, Mathilde Etcheverry, Arthur Gilly, Erwann Caillieux, Mohammed-Amin Madoui, Julie Guy, Amanda Bortolini Silveira, Stefan Engelen, Victoire Baillet, Patrick Wincker, Jean-Marc Aury and Vincent Colot

Institut de Biologie de l'Ecole Normale Supérieure (IBENS), Paris, France

Transposable elements (TEs) are mobile parasitic sequences that self-propagate and as a consequence many eukaryotic genomes are heavily laden with the remnants of once active TEs. Over evolutionary time, these TE remnants play an important role in generating new functions or the rewiring of gene regulatory networks. In contrast, the contribution of TE mobilization to the creation of heritable mutations remains unclear, partly because transposition is typically rare in nature, due to epigenetic repression and also because of the filtering effect of natural selection.

Using our population of *A. thaliana* epigenetic recombinant inbred lines (epiRILs), which are akin to mutation accumulation (MA) lines, we have characterized the creation of *de novo* TE-induced mutations in an essentially unbiased manner. We show that once initiated, transposition rapidly produces high loads of mutations and that three of the most active TE families in nature target specific non-overlapping subsets of genes with distinct chromatin features. Furthermore, we demonstrate that the histone variant H2A.Z directs the integration of *Ty1/Copia* retrotransposons into environmental response genes and away from essential genes, thereby maximizing the generation of epigenetically-sensitive mutations with adaptive potential. Overall, our findings reveal the non-random nature of TE-induced mutations and establish chromatin as a major determinant of their spectrum and phenotypic consequences.

Stable epigenetic inheritance of DNA methylation through pathway integration

Daniel Zilberman

John Innes Centre, UK

Cytosine DNA methylation is widespread among eukaryotes, including plants and animals. Dnmt1-family methyltransferases semi-conservatively copy methylation patterns following DNA replication by specifically methylating symmetrical CG dinucleotides. This ability to carry epigenetic information has been recruited by evolution for multiple functions, including transposon silencing and gene regulation. Proper methylation patterns are essential for mammalian and plant development and are disrupted in cancer. The semi-conservative model of DNA methylation inheritance is supported by much evidence and implies that the only stable states for CG sites within a population of cells should be fully methylated and fully unmethylated, because any maintenance inefficiency should eventually lead to full demethylation. Methylation patterns observed in diverse species are generally consistent with this expectation.

Our work with *Arabidopsis* plants deficient in two chromatin proteins, linker histone H1 and the nucleosome remodeler DDM1, revealed that intermediate levels of CG methylation can exist over a large fraction of the genome, and crucially that these can be stably inherited over many generations. We find that individual CG sites in these plants cycle rapidly between methylated and unmethylated states. We also find a similar behavior in wild type, but the cycling unfolds over multiple organismal generations instead of individual cell divisions. Our results indicate that the existing semi-conservative model is incomplete, and that stable inheritance of CG methylation is enabled by a balance between maintenance failures and untemplated *de novo* methylation. Our results therefore argue that truly stable epigenetic inheritance of DNA methylation is not a function of any individual system, but instead requires pathway integration.

Abstracts for the short talks

Epigenetic regulations in response to the environment

Iva Mozgová	p. 18
Clara Richet-Bourbousse	p. 19
Stéphane Maury	p. 20
Johan Zicola	p. 21
Clémentine Vitte	p. 22

Novel functions of PRC2 during embryo-to-seedling transition in *Arabidopsis*

Iva Mozgová¹⁻³, Helena Hönig Mondeková¹⁻³, Lenka Bučinská^{2,3}, Jiří Kubásek², Roman Sobotka^{2,3}, Jiří Šantrůček², Eliška Kuthanová Trsková^{2,3}, Aurelie Crepin³, Tomáš Konečný¹⁻³, Martin Tichý³, Kateřina Kabeláčová^{1,2}

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Embryo-to-seedling transition is a crucial developmental and metabolic phase transition in plants. In *Arabidopsis*, the transition is governed by the Polycomb Repressive Complexes (PRCs) that have a well-established role in ensuring stable transcriptional repression of key embryo maturation genes (Aichinger *et al.*, 2009; Bouyer *et al.*, 2011; Bratzel *et al.*, 2010; Chanvivattana *et al.*, 2004; Chen *et al.*, 2010).

We find that under standard photoautotrophic growth conditions, PRC2 is not necessarily needed for stable repression of embryonic genes, completion of photomorphogenesis and establishment of vegetative growth. Overcoming the requirement for PRC2 for the embryo-to-seedling transition, we study the function of PRC2 in the establishment and maintenance of photoautotrophic growth in plants, which has so far been hampered by the developmental arrest of plants fully depleted of PRC2 activity. Combining methodical approaches of developmental epigenetics, plant physiology and photosynthesis, we show that PRC2 moderates responses to ambient light. We aim to differentiate between biogenic (developmental) and operational processes governed by PRC2 in vegetative tissue. I will present our progress in identifying molecular pathways modulated by PRC2 with the goal to establish its function in vegetative tissue during photoautotrophic growth.

Aichinger, E., Villar, C.B., Farrona, S., Reyes, J.C., Hennig, L., and Kohler, C. (2009). CHD3 proteins and polycomb group proteins antagonistically determine cell identity in *Arabidopsis*. *PLoS Genet* 5, e1000605.

Bouyer, D., Roudier, F., Heese, M., Andersen, E.D., Gey, D., Nowack, M.K., Goodrich, J., Renou, J.P., Grini, P.E., Colot, V., *et al.* (2011). Polycomb repressive complex 2 controls the embryo-to-seedling phase transition. *PLoS Genet* 7, e1002014.

Bratzel, F., López-Torrejón, G., Koch, M., Del Pozo, J.C., and Calonje, M. (2010). Keeping cell identity in *Arabidopsis* requires PRC1 RING-finger homologs that catalyze H2A monoubiquitination. *Curr. Biol.* 20, 1853–1859.

Chanvivattana, Y., Bishopp, A., Schubert, D., Stock, C., Moon, Y.H., Sung, Z.R., and Goodrich, J. (2004). Interaction of Polycomb-group proteins controlling flowering in *Arabidopsis*. *Development* 131, 5263–5276.

Chen, D., Molitor, A., Liu, C., and Shen, W.-H. (2010). The *Arabidopsis* PRC1-like ring-finger proteins are necessary for repression of embryonic traits during vegetative growth. *Cell Res.* 20, 1332–1344.

Modeling the *Arabidopsis* DNA damage transcriptional response to identify new players in chromatin-based DNA repair mechanisms

Clara Bourbousse^{†1}, Neeraja Vegesna¹, Julie A. Law¹

¹ Plant Molecular and Cellular Biology Laboratory, Salk Institute for Biological Studies, La Jolla, CA, USA

[†] current address: Institut de biologie de l'Ecole normale supérieure (IBENS), Ecole normale supérieure, CNRS, INSERM, PSL Université, Paris, France

DNA damage produced from endogenous processes or exogenous sources must be recognized and repaired to prevent the accumulation of deleterious mutations. Double strand breaks (DSB) represent one of the most serious types of DNA damage as no intact strand remains as a template to direct repair. Upon detection of a DSB, a series of signaling cascades lead to the regulation of gene expression, cell cycle arrest or programmed cell death in severe cases, and finally to the orchestration of the repair process. Underscoring the importance of gene regulation in the DNA damage response, studies in *Arabidopsis* have demonstrated that all the aforementioned processes rely on SUPPRESSOR OF GAMMA RESPONSE 1 (SOG1), a NAC family transcription factor (TF) that has been functionally equated to the mammalian tumor suppressor p53. We generated transcriptional models of the DNA damage response from γ -irradiated seedlings during a 24-hour time course using DREM, the Dynamic Regulatory Events Miner, revealing 11 co-expressed gene clusters with distinct biological functions and *cis*-regulatory elements. Combining transcriptomic studies of the γ response in mutant backgrounds and ChIP analyses of TFs further revealed that SOG1 is the major activator, directly targeting the most strongly up-regulated genes, while Rep-MYB3R TFs are the major repressors, specifically targeting the most strongly down-regulated genes, which mainly correspond to G2/M cell cycle-regulated genes. In addition to providing information about transcriptional regulations, our network also has the potential to shed light on another aspect of the DNA damage response that is particularly under-studied in plants—the role of chromatin and its associated modifications in mediating DNA repair. Indeed, more than 50 chromatin-related genes (coding histone proteins, histone modifiers, readers or remodelers) were found in our DREM model, highlighting the importance of chromatin-based mechanisms in the DNA damage response. We further characterized one such chromatin reader and found that it affects DNA repair and may act specifically in shoot apical and flower meristems to maintain genome stability.

Epigenetic variation and plasticity in forest TREEs (EPITREE)

Maury Stéphane^{*a}, Fichot Régis^a, Sow Dia Mamadou^a, Delaunay Alain^a, Le Jan Isabelle^a, Rogier Odile^b, Ségura Vincent^b, Le Provost Grégoire^c, Ehrenmann François^c, Plomion Christophe^c, Salse Jérôme^d, Ambroise Christophe^e, Gribkova Svetlana^f, Tost Jorg^g, Mirouze Marie^h, Conde Danielⁱ, Allona Isabelⁱ, Strauss Steven^j

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Ongoing global climate changes in progress will impact forest productivity notably through reduced water availability and heat periods. One possibility to adapt is phenotypic plasticity for which epigenetic mechanisms are proposed to be a main source of flexibility. Our objective is to evaluate the potential of DNA methylation to significantly participate to phenotypic plasticity in trees, sessile and perennial organisms with major ecological roles. Over the 10 last years, using an integrative approach with ecophysiological, biochemical, transcriptomics, epigenomics (MeDIP, WGBS, Mobilome) and reverse genetics (RNAi lines) tools, we were able to dissect in the shoot apical meristem (center of the shoot morphogenesis) the response of trees to environmental variations. This work was assessed in distinct experimental set-ups from greenhouse to field plantations as well as during the stress or months post-stress. Our recent data^{1,2,3,4,5} showed that Differentially Methylated Regions (DMRs) are associated to active TE and differentially expressed genes with biological functions related to stress response and phytohormone signaling. Altogether, our data proposed that DNA methylation is a source of flexibility associated to phenotypic plasticity in trees opening perspectives for tree management. The role of epigenetic mechanisms in tree adaptation and microevolution will be also presented in the frame of the national project EPITREE 2018-2021 (ANR-17-CE32-0009-01, <https://www6.inra.fr/epitree-project/Le-projet-EPITREE>).

1 Conde D, Le Gac A-L, Perales M, Dervinis C, Kirst M, Maury S, González-Melendi P, Allona I (2017) Chilling-responsive DEMETER-LIKE DNA demethylase mediates in poplar bud break. *Plant Cell Environment* 40, 2236-2249. doi: 10.1111/pce.13019

2 Lafon-Placette C, Le Gac A-L, Chauveau D, Segura V, Delaunay A, Lesage-Descauses MC, Hummel I, Cohen D, Jesson B, Le Thiec D, Bogeat-Triboulot MB, Brignolas F, Maury S (2018) Changes in the epigenome and transcriptome of the poplar shoot apical meristem in response to water availability affect preferentially hormone pathways. *Journal of Experimental Botany* 69, 537-551 doi: 10.1093/jxb/erx409

3 Le Gac A-L, Lafon-Placette C, Chauveau D, Segura V, Delaunay A, Fichot R, Marron N, Le Jan I, Berthelot A, Bodineau G, Bastien J-C, Brignolas F, Maury S (2018) Winter-dormant shoot apical meristem in poplar trees shows environmental epigenetic memory. *Journal of Experimental Botany* 69, 4821-4837

4 Sow MD, Segura V, Chamailard S, Jorge V, Delaunay A, Lafon-Placette C, Fichot R, Faivre-Rampant P, Villar M, Brignolas F, Maury S (2018) Genetics and Genome Narrow-sense heritability and PST estimates of DNA methylation in three *Populus nigra* L. populations under contrasting water availability. *Tree Genetics & Genomes* 14

5 Sow, M. D. *et al.* (2018) Epigenetics in forest trees: state of the art and potential implications for breeding and management in a context of climate change. *Advances in Botanical Research* 88, 387-453.

Targeted DNA methylation represses two enhancers of *FLOWERING LOCUS T* in *Arabidopsis thaliana*

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FLOWERING LOCUS T (FT) plays a major role in regulating the floral transition in response to inductive long day (LD) photoperiod in *Arabidopsis thaliana*. Expression of *FT* in leaves is dependent on the distal transcriptional enhancer *Block C*, located 5 kb upstream of the transcriptional start site (TSS). We expressed an inverted repeat of *Block C* to induce local DNA methylation and heterochromatin formation, which lead to *FT* downregulation in inductive photoperiod. Using targeted DNA methylation as a tool to uncover additional regulatory regions at the *FT* locus, we identified *Block E*, located 1 kb downstream of the gene, as a novel enhancer of *FT*. Similarly to *Block C*, *Block E* is conserved across *Brassicaceae* and located in accessible chromatin. In combination with a minimal promoter, *Block E* drives phloem-specific expression of a reporter gene, indicating that *Block E* acts as transcriptional enhancer of *FT*.

Genome-wide hypermethylation of TEs and centromeres following low temperature exposure in maize

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Transposable elements (TEs) are major players in shaping genome structure. TE sequences are transcriptionally silenced by epigenomic modifications to limit the mutagenic potential of their transpositional activity. In particular, several DNA methylation pathways are responsible for TE silencing in the various chromosomal locations where TE reside. While DNA methylation is known to be modified by abiotic constraints, the extent to which it can be remodeled remains to be fully elucidated.

We show that low temperature triggers genome-wide hypermethylation in maize, mainly at transposable elements and centromeres. This hypermethylation is mediated by the parallel activation of multiple methylation pathways across chromosomes, to actively hypermethylate TEs in the various chromatin locations where they reside. This likely reflects the importance of taming transposable elements following an abiotic stress in maize, a species for which over 85% of the genome is constituted of transposable elements.

Functions of non-coding RNAs

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Plant transcriptome heterogeneity revealed by Transcript Isoform Sequencing (TIF-Seq)

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RNA polymerase II (RNAPII) transcription is co-transcriptionally linked to pre-RNA processing events that amplify the number of alternative transcripts isoforms from a single transcription unit (TU). To identify the scope of alternative transcript isoform production in plants, we developed transcript isoform sequencing (TIF-Seq) in *Arabidopsis*. TIF-seq maintains the information of the beginning and end of the same RNA molecule. We revealed unprecedented levels of TU boundary variation that expand proteomes by generating proteins with differing N-/C-terminal domain. We identify small promoter-proximal RNAs (sppRNAs) that overlap with transcription initiation regions of protein coding genes. sppRNAs appear to result from premature promoter-proximal transcriptional termination of a subset of genes. sppRNAs may be linked to the phenomenon of promoter-proximal pausing of RNAPII detected in many organisms. Our data highlight transient transcript isoforms associated with early stages of RNAPII transcription. Our TIF-seq data reveal plant transcript isoform diversity and inform on the magnitude of transcripts affected by genetic and epigenetic variation underlying plant fitness.

Dynamics of DNA methylation during symbiotic nodule development in *Medicago truncatula*

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The nitrogen-fixing endosymbiosis between legumes and rhizobia involves the formation of root nodules, which host rhizobia and provide them with a proper environment for nitrogen fixation and metabolic exchanges with the plant. Indeterminate nodules, such as those formed in the model legume *Medicago truncatula*, represent a very useful experimental system to investigate organogenetic and differentiation processes. Indeed, successive developmental stages coexist in a mature nodule, along a longitudinal axis from the apex (meristematic zone) to the basis of the nodule (nitrogen-fixing zone). The differentiation of both symbionts takes place in rhizobium-infected cells produced from the apical meristem, and involves the regulation of numerous plant and bacterial genes. Our goal is to investigate the mechanisms involved in the massive and coordinated upregulation of hundreds of plant genes in the differentiation zone. We recently showed that a substantial proportion (~35%) of these genes co-localize on the *M. truncatula* genome in >200 physical gene clusters, termed symbiotic islands (Pecrix et al., Nat Plants 2018).

A coupled laser-capture microdissection (LCM)-RNAseq approach allowed us to discover that the expression of genes involved in maintenance of DNA methylation is spatially regulated within the nodule. Those are strongly downregulated in the differentiation zone, while conversely the DNA demethylase DEMETER (DME) is strongly upregulated. This led us to analyze the importance of DME and the dynamics of DNA methylation during nodule development, using first selected genomic regions (Satgé et al., Nat Plants 2016). Latest results obtained with genome-wide Bs-seq analyses of whole organs (root tips and developing nodules) and laser-dissected nodule regions will also be presented.

Ecotype-related long non-coding RNAs in epigenetic regulation of root growth

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The plant root system is characterized by its high developmental plasticity. During phosphate starvation, root architecture varies widely between species and even between ecotypes of the same species despite the strong conservation of the protein-coding portion of their genomes. Using an RNAseq approach on two *Arabidopsis* ecotypes that have a contrasted response to phosphate starvation, we identified thousands of new lncRNAs. In contrast to protein coding genes that are highly conserved, non-coding RNAs evolved rapidly between ecotypes and may control their differential responses to the environment as several long non-coding RNAs (lncRNAs) can quantitatively regulate gene expression. We have identified 746 lncRNAs differentially expressed between the two ecotypes. Among them 345 co-localize with 24nt siRNAs suggesting a dynamic epigenetic status as these lncRNAs seems transcribed both by RNA pol II and RNA pol IV. Indeed, the APOLO lncRNA belonging to this class, regulates the formation of a chromatin loop, *via* RNA-dependent DNA methylation (RdDM) and active DNA demethylation, to control its neighboring gene. Two new lncRNAs were selected according to the deregulation of their expression in mutants affected in transcriptional gene silencing (TGS), their correlation of expression with neighboring genes in various conditions and their potential nuclear localization. Putative roles of these lncRNAs in epigenetic regulation are being explored through their mis-expression in root tissues and its consequences on *cis*-regulation of gene expression.

Differential epigenetic regulation of clustered nuclear tRNA genes expression in *Arabidopsis thaliana*

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Keywords: *Arabidopsis thaliana*, epigenetics, non-coding RNAs, tRNAs

The *Arabidopsis thaliana* nuclear genome contains around 600 tRNA genes (tDNAs). In general, each isoacceptor tRNA family owns a number of tDNA copies proportional to the corresponding codon usage frequency. However, Pro, Ser and Tyr tDNAs exhibit a surplus number of genes¹ organized in clusters of numerous repetitive elements. Whatever the case, these genes contain all required motifs for their transcription by the RNA polymerase III. Notwithstanding this, we recently revealed, using Northern blots and small RNA sequencing data analysis, that these clustered tDNAs are not expressed in normal growth conditions compared to the non-clusterised ones. Furthermore, using publicly available ChIP-seq and whole-genome bisulfite sequencing data, complemented by other molecular approaches, we unveiled particular differential epigenetic profiles at these clustered tDNAs. Based on these findings, our project aims at (i) deciphering the genetic and epigenetic determinants ruling the silencing of the clustered tDNAs, (ii) their possible reactivation in case of need following particular developmental or stress conditions and (iii) the functional impact of their particular epigenetic profiles on the organization of chromatin inside the nucleus. Together, our results highlight new features in the differential regulation of plant non-coding RNAs genes expression.

[1] Michaud, M., Cognat, V., Duchêne, A. M. & Maréchal-Drouard, L. A global picture of tRNA genes in plant genomes. *Plant J.* **66**, 80–93 (2011).

POST-TRANSCRIPTIONAL AND TRANSCRIPTIONAL REGULATIONS BY SMALL RNAS IN HEVEA LATEX CELLS

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Keywords: small RNA, miRNA, degradome, transposable element

In response to environmental cues and latex harvesting stress, *Hevea* revealed an alteration in sRNA transcriptome in latex cells associated with physiological disorders halting latex flow (Tapping Panel Dryness, TPD). Most sRNAs are 24 nt in healthy trees and 21 nt in TPD-affected trees. This questions the contribution of post-transcriptional gene silencing by sRNA to TPD. Firstly, *MIR* genes and miRNAs were annotated. Secondly, degradome data were used to highlight sRNA-mediated post-transcriptional genome expression regulation in response to abiotic stress and TPD. We sequenced all 3' ends of degraded mRNAs from 6 tissues, subjected or not to abiotic stress/TPD, in order to validate all sRNA-based target cleavage sites simultaneously. Our results showed that the 21-nt sRNAs accumulating during TPD were not derived from *MIR* genes and could therefore be classified as siRNAs, and correspond to epigenetically-activated siRNA (easiRNA). From “degradome” analysis from 6 distinct tissues, latex cells displayed the highest level of post-transcriptional regulation by mRNA cleavage. Interestingly, natural rubber biosynthetic pathway is under a strong post-transcriptional silencing. Genes involved in miRNA biogenesis were identified and the analysis of their sequences revealed a recent duplication of Argonaute (AGO) and Dicer-like (DCL) families in *Hevea* genome, consistent with the whole genome duplication shared with cassava. Partial conservation of miRNA-mediated post-transcriptional regulation was observed between *Hevea* and *Arabidopsis*. Discovery of miRNA/target couples through “degradome” analysis, annotation of *MIR* genes and transposable elements, representing more than 70% of the *Hevea* genome sequence and interspersed between coding genes, can lead to a full comprehensive picture of post-transcriptional and transcriptional regulations of gene expression by sRNA at genome-wide level.

Impact of DCL3 and RTL1 RNases in small RNA dynamics and epigenetic regulations in the nitrogen-fixing symbiosis of *Medicago truncatula*

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Small RNAs are essential regulators of gene expression during plant development, stress responses and plant-microbe interactions. These non-coding RNAs, 21-24 nucleotides long, down-regulate the expression of target genes through post-transcriptional (PTGS) or transcriptional (TGS) silencing. Several microRNAs, mainly targeting transcription factors, have been functionally associated to the control of symbiotic nodule development in legumes. However, much less is known about the roles of siRNAs in this process. Small RNAs are produced after cleavage of long double-stranded or hairpin RNA precursors by enzymes of the ribonuclease III family called DICER-LIKE proteins (DCL). DCL3 encodes the DCL involved in heterochromatic 24-nt sRNA biogenesis and RNA-directed DNA methylation (RdDM). In addition, an atypical RNase III-like, RTL1, was shown to inhibit siRNA production and act as a putative silencing repressor. Interestingly, a homolog of *RTL1* gene, *MtRTL1b*, is highly induced during symbiosis.

In our laboratory, we investigated the involvement of these two type III RNases in nodule development. In *Medicago truncatula* nodules, *MtDCL3* expression is higher in the meristematic zone than in the differentiation region of the nodule, suggesting a specific role of DCL3 and putatively 24nt siRNAs in nodule meristems. Furthermore, we showed that mutations (a Tnt1 insertion) or *MtDCL3*RNAi lines of developed bigger, multi-lobed nodules. In contrast, RNAi of the nodule-specific gene *MtRTL1b* led to impaired nodule development, reduced nitrogen fixation and increased bacteroid lethality. The impact of these mutations on small RNA dynamics, notably 24nt siRNAs, is ongoing. We propose that these RNAseq linked to siRNAs are new regulators of the *Medicago* symbiosis.

Chromatin Dynamics

Christophe Laloi	p. 31
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Topoisomerase VI participates in an insulator-like function that prevents heterochromatin spreading into euchromatin islands

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The organization of the genome into transcriptionally active and inactive chromatin domains requires well-delineated chromatin boundaries and insulator functions in order to maintain the identity of adjacent genomic loci with antagonistic chromatin marks and functionality. In plants that lack known chromatin insulators, the mechanisms that prevent interference between adjacent chromatin domains and the spreading of heterochromatin into euchromatin remain to be identified. Here, we show that DNA Topoisomerase VI participates in a chromatin boundary function that safeguards the expression of genes in euchromatin islands within silenced heterochromatin regions. While many transposable elements are reactivated in mutants of the Topoisomerase VI complex, genes insulated in euchromatin islands within the large heterochromatic regions of the *Arabidopsis thaliana* genome, namely the pericentromeric and knob regions, are specifically downregulated. H3K9me2 levels consistently increase at euchromatin island loci and decrease at some TE loci. We further show that Topoisomerase VI physically interacts with S-adenosylmethionine (SAM) synthase MAT3, which is required for H3K9me2 deposition. Topoisomerase VI promotes MAT3 occupancy on heterochromatic elements and its exclusion from euchromatic islands, thereby providing a mechanistic insight into the essential role of Topoisomerase VI in the delimitation of chromatin domains. This study unveils the existence of an insulator-like function in *Arabidopsis thaliana* that requires Topoisomerase VI to prevent heterochromatin spreading into nearby euchromatic islands.

Histone H3.3 Deposition in *A. thaliana*

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Histones are essential components of the nucleosome, the basic subunit of chromatin that structures linear DNA molecules and regulates access of other proteins to DNA. DNA accessibility can be modulated through specific post- translational modifications of histones or the incorporation of different variants of the core histones H3, H2A and H2B into the nucleosome. A network of histone chaperone complexes, highly conserved through evolution, ensures the well-timed deposition of these variants. The histone variant H3.1 for example is incorporated during DNA replication, while the variant H3.3 is deposited in a DNA synthesis- independent manner.

To better understand how the deposition of H3.3 variants is controlled in plants, we have characterized the *Arabidopsis* histone chaperone complexes Histone Regulator A (HIRA) and Alpha Thalassemia-mental Retardation X-linked (ATRX) and show that loss of these complexes affects nucleosomal occupancy and gene expression. *Arabidopsis* HIRA and ATRX mutant alleles are viable, but cause severe developmental defects when combined together, suggesting that HIRA and ATRX function in complementary histone H3.3 deposition pathways. Indeed, *Arabidopsis* ATRX binds H3.3 and ATRX loss-of-function reduces cellular histone H3.3 pools and in consequence modulates the H3.1/H3.3 balance in the cell. At the genome-wide scale, our data indicate that ATRX modifies gene expression through H3.3 deposition at a set of genes characterized both by elevated H3.3 occupancy and high expression levels, altogether emphasizing the role of histone chaperones in regulating chromatin dynamics and fine-tuning genome expression.

Chromosome architecture of hexaploid wheat (*Triticum aestivum* L.)

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Polyploidization events are known to trigger extensive epigenetic and transcriptional alteration of the duplicated or merged genomes, accompanied by small- and large-scale conformational changes. The genome of modern hexaploid wheat (*Triticum aestivum* L.; $2n = 6x = 42$) is the product of two rounds of interspecific hybridization between three closely related diploid species, resulting in the presence of distinct but highly syntenic sub-genomes (AA, BB and DD). We examined the large-scale chromatin architecture of the nucleus of wheat using Hi-C, a genome-wide chromatin conformation capture (3C) method and GISH, (genomic *in situ* hybridization). We found evidence that physical interactions occur with significantly higher frequency within sub genomes (A with A, B with B or D with D) than between sub genomes (A with B or D, etc. ...), defining sub-nuclear “genomic territories”. In addition, all chromosomes are folded into a V-shaped conformation that brings the gene-rich subtelomeric regions in close proximity. This organization determines a polarized distribution of euchromatin and heterochromatin that indicates a functional compartmentalization within the nucleus. On a local scale, we found that genes tend to interact mainly with other genes over long-distance “loops”, and especially with genes presenting similar expression levels and bearing the same histone marks. Moreover, gene pairs involved in these chromatin loops show similar changes in expression levels between shoots and roots. This suggests that local-scale topology is an important factor for transcriptional regulation and may reflect the presence of “transcription factories” in wheat. Our results provide a framework for further understanding the physical organization of wheat genome and highlight the relationship between epigenetic marks and chromosome conformation and how this affects gene expression.

Regulation of chromatin mark changes for organogenesis in *Arabidopsis*

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Plant life-long organogenesis involves sequential, time and tissue specific expression of developmental genes. This requires changes in chromatin marks, particularly those deposited at Histone 3 by the repressive Polycomb Group (PcG) PRC2 and activating trithorax Group (trxG) complexes. While genome-wide profiles of PRC2-induced H3K27me3 and trxG-induced H3K4me3 modifications have been reported in plants for several tissues or conditions, only few studies, including ours, have depicted their dynamics during specific developmental processes. The mechanisms that switch the chromatin states of genes (in particular from repressed to active) during organogenesis are even more elusive. We use a combination of genome-wide, genetic and molecular interaction analyses to elucidate these mechanisms in *Arabidopsis*. I will report our advances on the functions and interactions of antagonists of the PRC2 CURLY LEAF H3K27me3 methyl transferase: the *jumonji* H3K27me3 demethylases and the chromatin state switch ULTRAPETALA1 (ULT1). I'll also mention our recent data on ULT1 complex components, and how it reveals intricate connections between repressive and activating chromatin factors.

A transgene locus mimics transposon *de novo* silencing

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Transcriptional gene silencing (TGS) controls the expression of transposable elements and of endogenous genes bearing repeats in their promoter, and is associated with increased DNA methylation. We previously described a transcriptionally silenced and methylated transgene locus, named *L5*, which consists of six to seven T-DNA repeats carrying a *pNos-nptII* selectable marker and a *p35S-GUS* reporter gene. We and others have shown that *L5* locus is strongly reactivated in *ddm1* and *met1* mutants and to a lesser extent in *main*, *mail*, *mom1*, *bru1*, *hda6*, *hog1*, *mom1*, *rpa2*, *cmt3*, *drm2*, *fas1*, and *fas2* mutants. In contrast, mutations in the RNA-directed DNA methylation (RdDM) pathway do not affect *L5* silencing. Nevertheless, *L5* acts in *trans* to silence transgenes driven by a 35S promoter but not transgenes expressing *GUS* under a different promoter. Such *trans*-silencing is impaired in RdDM mutants, indicating that the RdDM pathway is dispensable for maintenance of TGS at the *L5* locus but is required for *de novo* establishment of TGS in *trans*. Given this result, we transiently removed DDM1 to reactivate *L5* and followed the process of *de novo* establishment of *L5 cis*-TGS in various mutant back-grounds. Our results show that the production of RdDM-dependent siRNAs is required for *de novo* establishment of *L5 cis*-TGS. Whether RDR6 also contributes to the establishment of RdDM at the *L5* locus is under investigation.

The NRPD1 N-terminus is critical for Pol IV-based genome surveillance in *Arabidopsis*

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Genome surveillance pathways in eukaryotes help limit mutations and other deleterious effects of transposable elements (TEs). Plants use a specialized non-coding RNA machinery, including RNA polymerase IV (Pol IV), to ensure this selective silencing of TEs. Pol IV and its physically coupled partner, RDR2, synthesize ~30bp double stranded RNAs (dsRNAs) from thousands of distinct chromosomal loci, most of which are TEs. These dsRNAs are processed into 24 nt small interfering RNAs (siRNAs), which guide DNA methylation and silencing to related TE targets. Pol IV is a twelve-subunit enzyme that evolved as a specialized form of RNA polymerase II (Pol II). The largest subunit of Pol IV, NRPD1, is hypothesized to contain key determinants of the enzyme's unique functions. Here we used molecular genetic approaches to study the role of the NRPD1 N-terminus in TE methylation and surveillance. *Arabidopsis* lines harboring missense mutations in this N-terminus produce wild-type levels of NRPD1, which co-purifies with other Pol IV subunits and RDR2. Our *in vitro* transcription, small RNA-seq and methyl-seq analyses reveal that the NRPD1 N-terminus is critical for robust Pol IV-dependent transcription, siRNA production and DNA methylation at most chromosomal targets. Residual DNA methylation observed in one N-terminus mutant, however, indicates that Pol IV function can be partially uncoupled from the elevated siRNA levels observed in wild-type plants. This mutation crippled Pol IV's ability to inhibit retrotransposon activation under heat stress. Based on these results and differences in amino acid conservation, we propose that the NRPD1 N-terminus diverged from the related NRPB1 (Pol II) subunit to support Pol IV function in genome surveillance.

Transgenerational epigenetic variation

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Role of Plant Mobile Domain proteins in regulation of gene expression

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Plant mobile domain (PMD) proteins are related to genome stability, developmental processes, and silencing of transposable elements (TEs) (Wenig *et al.*, 2013; Ühlken *et al.*, 2014; Ikeda *et al.*, 2017). In addition, it was initially proposed that PMDs could act as transcription factors (TFs) derived from TEs after gene domestication (Babu *et al.*, 2006; Steinbauerova *et al.*, 2011). Using a forward genetic screen, we recently identified MAINTENANCE OF MERISTEMS (MAIN), the founding member of PMD family in *Arabidopsis thaliana*, as a protein required for TE silencing and proper gene expression. DNA methylation is one of the many pathways that converge towards the silencing of TEs, and that can regulate gene expression. Among other enzymes, DNA methylation is mediated by DRM1, DRM2 and CMT3 (Law and Jacobsen, 2011). We performed RNA sequencing (RNA-seq) analyses in the *drm1 drm2 cmt3 main-3* (*ddc main-3*) quadruple mutant in comparison to the *ddc triple*, and the *main-3* hypomorphic single mutant plants. These analyses showed a synergistic effect of the DRM2, CMT3 and MAIN pathways in TE silencing and regulation of gene expression. RNA-seq experiments using the *main-2* null allele confirmed the TE silencing defects and misregulation of gene expression observed in *main-3*. We also performed transcriptomic analyses in the *main-like1-1* (*mail1-1*) null mutant, which revealed significant overlaps of misregulated loci between the two *pmd* mutants. Considering that *main-2* and *mail1-1* display similar developmental phenotypes, we postulated that MAIN and MAIL1 may physically interact together, which was confirmed by co-immunoprecipitation experiments. Interestingly, among the genes that are downregulated in the two *pmd* mutants, we identified *Microrchidia1* (*MORC1*), known for its role in TE silencing (Moissiard *et al.*, 2012, 2014). Thus, the downregulation of *MORC1* could explain the TE upregulation observed in the two *pmd* mutants. Finally, analyzing the chromatin states of genes that are upregulated in the *pmd* mutants showed an enrichment for Polycomb-targeted genes. We hypothesize that MAIN and MAIL1 may act as TFs at specific loci, such as the *MORC1* locus, and that the upregulation of TEs observed in the *pmd* mutants could be an indirect effect.

Babu, M.M., Iyer, L.M., Balaji, S., Aravind, L. (2006). The natural history of the WRKY-GCM1 zinc fingers and the relationship between transcription factors and transposons. *Nucleic Acids Research* 34, 6505-6520.

Ikeda, Y., Pélissier T., Bourguet, P., Becker, C., Pouch-Pélissier, M.N., Pogorelnik, R., Weingartner, M., Weigel, D., Deragon, J.M., Mathieu, O. (2017). *Arabidopsis* proteins with a transposon-related domain act in gene silencing. *Nature Communication* 8, 1-10.

Law, J. A., Jacobsen, S. E. (2011) Establishing, maintaining and modifying DNA methylation patterns in plants and animals. *Nat. Rev. Genet.* 11, 204-220.

Moissiard, G., Cokus, S.J., Cary, J., Feng, S., Billi, A.C., Stroud, H., Husmann, D., Zhan, Y., Lajoie, B.R., McCord, R.P., Hale, C.J., Feng, W., Michaels, S.D., Frand, A.R., Pellegrini, M., Dekker, J., Kim, J.K., Jacobsen, S.E. (2012) MORC Family ATPases required for Heterochromatin condensation and gene silencing. *Science* 336, 1448-1451.

Moissiard, G., Bischof, S., Husmann, D., Pastor, W.A., Hale, C.J., Yen, L., Stroud, H., Papikian, A., Vashisht, A.A., Wohlschlegel, J.A., Jacobsen, S.E. (2014) Transcriptional gene silencing by *Arabidopsis* microrchidia homologues involves the formation of heteromers. *PNAS* 111, 7474-7479.

Steinbauerova, V., Neumann, P., Novak, P., Macas, J. (2011) A widespread occurrence of extra open reading frames in plant Ty3/gypsy retrotransposons. *Genetica* 139, 1543-1555.

Ühlken, C., Horvath, B., Stadler, R., Sauer, N., Weingartner, M. (2014). MAIN-LIKE1 is a crucial factor for correct cell division and differentiation in *Arabidopsis thaliana*. *The Plant Journal* 78, 107-120

Wenig, U., Meyer, S., Stadler, R., Fischer, S., Werner, D., Lauter, A., Melzer, M., Hoth, S., Weingartner, M., Sauer, N. (2013). Identification of MAIN, a factor involved in genome stability in the meristems of *Arabidopsis thaliana*. *The Plant Journal* 75, 469-483.

Pol epsilon links DNA replication with gene silencing, heterochromatin structure and DNA methylation

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Transcriptional gene silencing is maintained by multiple layers of repressive chromatin marks, such as DNA methylation or the H3K9me2 histone mark. In a genetic screen designed to identify transcriptional silencing factors, we isolated a new hypomorphic mutant allele of *POL2A*. *POL2A* encodes the catalytic subunit of the DNA polymerase epsilon, responsible for leading strand elongation during DNA replication, and known to maintain silencing at some pericentromeric repeats. Profiling transcriptome and repressive chromatin marks, we show that silencing defects in *pol2a* mutants do not correlate with decreases in DNA methylation or in the repressive histone marks H3K9me2, H3K27me1 and H3K27me3. However, we find that heterochromatin integrity is strongly perturbed in mutant *pol2a* nuclei.

Unexpectedly, *pol2a* mutations induce DNA hypermethylation in the CHG and CHH contexts, accompanied by H3K9me2 overaccumulation. While our data shows that CMT3 overexpression is likely responsible for a large part of DNA hypermethylation in *pol2a* mutants, further analysis also suggests that another mechanism is at play. This second mechanism appears to be linked with DNA replication hindrance, suggesting that replication stress participates to the molecular phenotypes of *pol2a* mutants.

Our study indicates that POL2A maintains transcriptional silencing, possibly by promoting heterochromatin structure, but also inhibits DNA methylation at non-CG sites by preventing CMT3 overexpression and replication stress.

***Arabidopsis* natural and induced epialleles associated with quantitative resistance to clubroot**

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Quantitative disease resistance, often influenced by environmental factors, is thought to be the result of DNA sequence variants segregating at multiple loci. However, heritable differences in DNA methylation, so-called transgenerational epigenetic variants, could also contribute to quantitative traits. Clubroot caused by the protist *Plasmodiophora brassicae* is a major disease of *Brassicaceae* including the three most economically important *Brassica* species: *B. napus*, *B. rapa*, *B. oleracea* and the model plant *Arabidopsis thaliana*. Both qualitative (major genes) and quantitative (QTL) clubroot resistance have been identified and studied in *Brassicaceae*.

Here we first describe a natural epimutation controlling a moderate-effect QTL involved in *Arabidopsis* clubroot resistance. Fine mapping of the QTL, previously identified in a biparental population from the cross between a susceptible and a partially resistant accession, was first carried out using 2400 progenies from Heterogeneous Inbred Families. We identified a 25kb-region containing eight genes with a reduced number of SNP between the parental accessions. Transcriptomic and methylation data analyses then highlighted in this region a causal 6kb epimutation spreading two genes. Significant correlations found between high methylation levels, gene repression of both genes and susceptibility to clubroot were lastly revealed in 127 natural *Arabidopsis* accessions, validating the implication of a natural epimutation in the variation of plant response to clubroot infection.

To determine to what extent epimutations could be used as diversity source in clubroot resistance, we then used the epigenetic recombinant inbred line population (epiRIL) derived from the cross *ddm1-2* x Col-0, which shows extensive epigenetic variation but limited DNA sequence variation. Quantitative loci under epigenetic control (QTL^{epi}) mapping was carried out on 123 epiRIL infected with *P. brassicae* and using various disease-related traits. EpiRIL displayed a wide range of continuous phenotypic responses. Twenty QTL^{epi} were detected across the five chromosomes, with *a bona fide* epigenetic origin for sixteen of them. The effect of three QTL^{epi} was dependent on temperature conditions. Six QTL^{epi} co-localized with previously identified clubroot resistance genes and QTL in *Arabidopsis*.

These results revealed the implication of natural and induced epimutations in the variation of plant response to clubroot infection. Combination of favorable epiallelic variations to allelic ones could therefore constitute new opportunities to breed plant resistant varieties to biotic stress.

AT-hook proteins increase histone modification H3K9me2 independently of DNA methylation

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Matrix attachment regions (MARs) are important for chromatin organization and gene expression. MARs are stretches of AT-rich sequences that guide binding of DNA to the nuclear matrix by recruiting MAR-binding proteins. Proteins with AT-hook motifs bind to MARs and affect the epigenetic state of target regions in animals and plants. Some AT-hook proteins are known to regulate gene expression in plants, but their functional mechanism is largely unknown. We have identified an AT-hook protein AHL10 and the SET domain-containing SU(VAR)3–9 homolog SUVH9 as interacting partners of ADMETOS (ADM). *ADM* is a paternally expressed imprinted gene that builds a hybridization barrier when *Arabidopsis* species of different ploidy hybridize. Significantly increased expression of *ADM* in *Arabidopsis* triploid seeds results in H3K9me2 hypermethylation in MARs. H3K9me2 hypermethylation in MARs was abolished in mutants of *adm* and ADM-interaction partners. Further analysis revealed that AHL10-mediated H3K9me2 hypermethylation in MARs is independent of DNA methylation. Moreover, we find that another AT-hook protein, AHL22, is capable of increasing H3K9me2 in sporophytic tissues, independent of DNA methylation. Our data suggests a novel mechanism of DNA methylation-independent H3K9me2 deposition that acts on MARs.

A novel RdDM-based reporter system to dissect transgenerational epigenetic inheritance in *Arabidopsis*

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In plants, RNA-directed DNA methylation (RdDM) is a key epigenetic process that triggers DNA methylation through small interfering RNAs (siRNAs). Although the molecular basis of RdDM establishment, maintenance and removal have been extensively studied in plants, very little is known about the mechanisms that control siRNA-directed transmission of epigenetic states. We have generated a reporter system to monitor such epigenetic transmission in the model plant *Arabidopsis thaliana*. This sensor, namely *IR-CHL*, expresses an inverted repeat targeting a specific promoter region of a gene involved in chlorophyll biosynthesis, named here *CHL*. Constitutive expression of the *IR-CHL* hairpin in *A. thaliana* results in primary transformants (T₁) having leaves with variegated pattern of chlorosis caused by *IR-CHL* mediated gene silencing activity at the targeted promoter region. Both siRNAs and DNA methylation highly accumulated in the T₁ Col-0 plants carrying *35S::IR-CHL*. More specifically, we found that siRNAs and non-CG mark were elevated in chlorotic sectors compared to green sectors of T₁ plants, indicating that the accumulation of siRNAs and of DNA methylation, must reach a certain threshold to initiate the variegated chlorotic phenotype. Particularly, we observed that siRNA accumulation was enhanced in one of the two DNA strands of the *CHL* promoter target, which is potentially transmitted clonally through mitosis, resulting in chlorotic sectors as observed in classical maize and tomato natural epialleles. Expression of *35S::IR-CHL* in mutants defective in specific RdDM components unveiled the genetic requirement of this reporter system and implicated a major role of RNA polymerase V for the transcriptional gene silencing of *CHL*. Significantly, the variegated phenotype in the primary transformants was found to be inherited to the next generation in a small RNA and DNA methylation dose-dependent manner. Here I will present the characterization of this novel RdDM reporter. I will also discuss how this system can be exploited to identify (i) novel RdDM factors and (ii) components that control siRNA-directed transgenerational inheritance of epigenetic states.

Epigenetic reprogramming associated with wound-induced organ regeneration

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During normal development, a vast rearrangement of the epigenetic landscape is correlated with the progression from undifferentiated cells into differentiated cells. This epigenetic reprogramming serves as a barrier, preventing cells to regress from a differentiated state back into an undifferentiated state. Interestingly, plants can reactivate cell proliferation after wounding to heal wounds and regenerate organs or even whole plants. This means cells at the wound site can dedifferentiate and overcome the epigenetic barrier preventing this. We characterized the rearrangement of the epigenetic landscape at different time points after wounding by ChIP-seq for several histone modifications (H3K4me3, H3K36me, H3K27me3, H3K9/14ac, H3K27ac) in combination with a concurrent genome-wide transcript profiling. These global analyses indicate genes rapidly induced by wounding are associated with elevated H3K9/14ac and H3K27ac levels before and/or right after wounding. Moreover, chemical intervention of GNAT-MYST-mediated histone acetylation strongly blocks wound-induced transcriptional activation as well as callogenesis at wound sites. This study thus highlights a prominent role for histone acetylation in plants capacity to regenerate.

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ULTRAPETALA1 antagonizes Polycomb PRC2 function independently of the histone demethylase REF6 in *A. thaliana*

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The switches in gene expression that occur during the development of pluricellular organisms involves Polycomb Group (PcG) proteins and Trithorax group (TrxG) proteins that respectively silence and activate gene expression *via* chromatin editing. Little is known about how activities of these proteins are regulated in a context-dependent manner. In *Arabidopsis thaliana*, developmental genes such as floral homeotic genes are associated with Histone 3 Lysine 27 trimethylation (H3K27me3) repressive epigenetic marks, which prevents the expression of genes out of their proper developmental context. These marks are deposited and maintained by the Polycomb Repressive Complex 2 (PRC2) that contains the histone methyltransferase CURLY LEAF (CLF). The SAND domain protein ULTRAPETALA1 (ULT1) antagonizes CLF by reducing H3K27me3 levels on its target genes, allowing their expression at the appropriate time and location. ULTRAPETALA1 molecular mode of action is still unknown. It could actively cause removal of H3K27 trimethylation marks at targets *via* an interaction with a protein carrying a demethylase activity such as RELATIVE OF FLOWERING 6 (REF6), and/or prevent *de novo* deposition of H3K27me3 *via* an interaction with Polycomb Repressive Complex 2. We show that REF6 and ULT1 act in independent pathways, suggesting that ULT1, rather than promoting active removal of histone marks, may prevent their *de novo* deposition.

PRC2 regulates root apical meristem homeostasis in *Arabidopsis thaliana*

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Polycomb Repressive Complexes 2 (PRC2) play a major role in maintaining the transcriptional repression of target genes by regulating their chromatin accessibility *via* the trimethylation of histone H3 lysine 27. We use the *Arabidopsis* root tip as a model to investigate the role of PRC2 in the regulation of cellular transitions during organogenesis. Previous results showed that PRC2 activity is important to maintain cellular identity upon specification. Recent observations indicate that PRC2 activity is critical to maintain the quiescent center (QC) population within the stem cell niche (SCN) and to regulate the timing of differentiation. Characterization of the PRC2 targets dynamically regulated within the root tip identified regulators of the stem cell niche as well as differentiation factors. Our current efforts aim at establishing the functional impact of PRC2-mediated gene repression in coordinating the gene regulatory networks underlying SCN homeostasis.

Synergistic interactions between miRNAs and Polycomb-Group proteins for the regulation of the CUC2 gene expression during leaf development

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Polycomb-Repressing Complex 2 (PRC2)-mediated chromatin remodelling and miRNA-mediated regulation are two epigenetic mechanisms regulating gene expression respectively at the transcriptional and post-transcriptional levels. How these two levels of regulation may interact is not well known. *CUP-SHAPED COTYLEDON 2* (*CUC2*), a gene quantitatively controlling *Arabidopsis* leaf serration, harbours repressive histone marks deposited by the PRC2 and is regulated by the miR164 which is encoded by 3 genes in *Arabidopsis*, *MIR164A*, *B* and *C*. Here we used *CUC2*-controlled leaf shaping as a model to precisely analyse the relative contributions and the interactions between miRNA and Pc-G proteins-mediated gene regulation. We show that *MIR164A* is a direct target of the *CUC2* transcription factor and creates a negative feedback circuit regulating *CUC2* protein levels but not its spatial nor temporal pattern. Additionally, we show that the PRC2 represses *CUC2* transcription, but surprisingly, this is not translated into an increased *CUC2* protein level as revealed by both *CUC2* protein *in situ* quantification and leaf morphometrics. We further provide evidence that this discrepancy between *CUC2* transcription and protein accumulation in PRC2 mutants is due to the emergence of a new regulatory interaction between *CUC2* and *MIR164B* that is not active in a background with wild-type PRC2 activity. This work exemplifies how defective transcriptional regulations can be compensated by the activation of a new post-transcriptional regulation and illustrates how the synergistic interactions between two fundamental mechanisms of gene expression regulation contribute to maintain robust development despite flexible underlying genetic circuits.

Heterochromatin reshaping upon UV exposure

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According to their sessile life style and to sunlight dependency, plants need to efficiently respond to light stress. Indeed, UV induce developmental responses but also DNA lesions that may affect genome and epigenome integrities. Recent studies performed in our team, showed that UV irradiation induce significant alterations of DNA methylation levels at centromeric/pericentromeric regions. Such changes in methylation landscape are over amplified in several DNA repair deficient plants. In order to determine whether such changes are accompanied by histones modifications and heterochromatin reshaping we analyzed nuclei of *Arabidopsis* roots before and 24h after UV exposure, using DAPI staining and immunolabelling of two repressive marks of constitutive heterochromatin: H3K9me2 and H3K27me1. We observed, differences in the intensity, size and shape of the heterochromatic signals, suggesting a remodeling of heterochromatin as response to UV stress. To perform our analyses, we have developed a pipeline linked to a semi-automatic program: “NuclEye”, allowing quantification of the signals and size and shape analyses. The results of these analyses open the discussion about the role of different DNA repair factors in the formation or maintenance of constitutive heterochromatin architecture and composition upon UV exposure.

Heritability of epigenetic marks and its impact on phenotypic variability in apple

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Because of its auto-incompatibility, apple is an obligate outcrosser and highly heterozygous. To maintain traits, apple trees are grafted thereby ensuring the stability of these varieties. The Golden Delicious variety was initially identified over 120 years ago and has been grafted ever since. Due to this clonal propagation, the Golden Delicious genome of today is mostly identical to the initial pippin that was initially obtained. Yet, phenotypic variability in clonally propagated apple is apparent, suggesting that epigenetic mechanisms may contribute to that variability. However, epigenetic transmission mechanisms are not yet well known in perennial plants. Moreover, the plasticity of the epigenome over time may allow adaptation to environmental conditions and pathogen attacks. Therefore, we can expect that the current apple epigenome differs from the original progenitor. To explore this question, we defined two main lines of research: (i) Because DNA methylation is the hallmark of epigenetic marks, we wanted to better understand the importance of this mark in apple development using the CRISPR-Cas9 technology: we expect to induce global reduction of DNA methylation by a targeted knockout of the apple homologues of the DNA methylation maintenance enzyme MET1 of *Arabidopsis*. (ii) In order to assess the heritability of epigenetic marks during sexual and asexual reproduction in apple, we compare the epigenomes of donor-trees, grafts (asexual) and seedlings obtained from self-fertilization (sexual). Here, as a model variety we use our Golden Delicious double haploid line, which greatly simplifies downstream bioinformatic analyses. First results suggest numerous differentially methylated regions (DMRs) between each of the analyzed samples. Interestingly, there are more DMRs between seed and graft or tree compared to tree and graft suggesting that tree and graft are more closely related at the epigenetic level. To understand the impact of these DMRs on gene expression we will carry out transcriptomic and phenotyping analyses. This work will improve our understanding of the contribution of epigenetics to the phenotypic diversity that is observed within an apple genotype.

Control of *EVD* and other *COPIA* elements silencing in *Arabidopsis thaliana*

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Transposable elements (TEs) are major components of plant genomes. Their capacity to travel to distant loci not only makes them a threat to genome integrity but also a tool for evolution. Their expression is, therefore, tightly regulated by different mechanisms, one of which is DNA methylation, in particular CG methylation. A null mutation in the *METHYLTRANSFERASE1* (*MET1*) gene causes a general loss of CG methylation associated with a reactivation of many TEs. The *Evadé* (*EVD*) retrotransposon is also reactivated but only after floral transition. It was also observed that transposition of *EVD* only occurs after several generations of inbreeding. These observations suggest that there are different levels of silencing that control *EVD* activation and transposition. To explore these different layers of silencing we are generating successive generations from a *met1-7* heterozygous plant in which we are studying the reactivation and the chromatin context of *EVD* and other *COPIA* elements. In parallel, we are producing a line of backcrossed *met1-7* plants in order to examine the effects of inbreeding in the control of TE transposition. Analysing the first generation of the segregating *met1-7* mutant allowed us to confirm the specific reactivation pattern of *EVD* and, in a lesser extent, of two other *COPIA* elements, *COPIA27* and *COPIA48*. Surprisingly, our results also show a reactivation of *COPIA27* and *COPIA48* in wild type and heterozygous plants obtained from the segregation of the *met1-7* heterozygous plants.

Modification of DNA Methylation

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DNA methylation, in association with other epigenetic marks such as histone modification, regulates chromatin structure and gene expression. In eukaryotes, DNA methylation is mediated by the addition of a methyl group obtained from S-adenosylmethionone (SAM) to carbon 5 of a cytosine residue. In plants, it involves *de novo* and maintenance of methylation in CG, CHG and CHH (H representing A, C or T) contexts, catalysed by three classes of methyltransferases, namely methyltransferase 1 (MET1), chromomethyltransferase 3 (CMT3), and domains rearranged methyltransferase 2 (DRM2). *De novo* methylation in all sequence contexts is carried out by DRM2 as part of the RNA-directed DNA methylation (RdDM) pathway, while maintenance of CG and CHG methylation is catalysed by MET1 and CMT3. In the RdDM-independent DNA methylation pathway, MET1 plays a central role in regulating cytosine methylation marks in all sequence contexts in which MET1 may co-ordinate the formation of methylation complexes. We aim to investigate how by over- expressing different forms of *MET1*, with and without catalytic site and SAM binding domain can alter epigenetic states at target genes controlled by the RdDM-independent methylation pathway and therefore cause epigenetic variation in plants. Phenotypic analysis of the *Arabidopsis* transformants of *MET1* with and without catalytic site (namely *METo-A* and *METo-I* lines) revealed reduction in primary root length in all lines and delayed germination in some of the line. Further analysis of germination assays with hormone treatments such as IAA, kinetin, ABA, GA and inhibitor paclobutrazol suggested that the hormonal imbalance between ABA and GA levels might be causal for delayed germination in the *METo-A* line.

In contrast, *Arabidopsis* transformants of *MET1* without catalytic site and SAM binding domain (namely *MSM* line) lack the phenotypes that were observed in the *METo* lines. Our results suggest the requirement of the SAM binding domain of *MET1* for the phenotypic effects produced by *MET1* over-expression.

Regulation of PAMP-induced transcriptional reprogramming by the active DNA demethylase ROS1

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DNA methylation is an epigenetic mark that silences transposable elements as well as some genes carrying repeats in their vicinity. In *Arabidopsis*, CG and CHG methylation marks are maintained by MET1 and CMT3, respectively, while CHH methylation is maintained by the RNA-directed DNA methylation (RdDM) machinery. Furthermore, the demethylase REPRESSOR OF SILENCING1 (ROS1) actively erases DNA methylation in all cytosine sequence contexts. We have previously shown that MET1 and RdDM factors negatively regulate resistance against a virulent strain of *Pseudomonas syringae*, and accordingly, siRNAs and DNA methylation were found to repress the expression of defense genes including functional resistance (*R*) genes. By contrast, we reported that *ROS1* positively regulates plant defense against this bacterium and have identified a *R* gene that is directly controlled by this active DNA demethylase. By using whole genome mRNA-, small RNA- and bisulfite-sequencing analyses, we have retrieved the whole set of immune-responsive genes that are controlled by ROS1 upon elicitation with flg22, a classical Pattern Associated Molecular Pattern (PAMP) derived from the N-terminal part of flagellin. These include not only canonical *R* genes but also membrane-associated receptors which could be potential PAMP receptors and several uncharacterized genes. In-depth characterization of the regulation at several candidates indicates that active demethylation at their promoters is critical for their flg22-induced transcriptional activation. Importantly, we show that hypermethylation in *ros1* mutants is mainly driven by DCL3- and DCL2-dependent siRNA and that these siRNA are crucial for the hypersusceptibility phenotype observed in *ros1* mutant. By analyzing regions hypermethylated in *ros1* at candidate loci, we have identified transcription factors (TF) binding sites suggesting that active demethylation by ROS1 might function by facilitating TF binding at the border of promoter localized TEs and repeated elements.

Effect of latitude, ecotype and zinc pollution on DNA methylation in *Noccaea caerulescens*

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Noccaea caerulescens is a plant species from the family of *Brassicaceae* that can tolerate and accumulate high levels of heavy metals in soil, at concentrations that are toxic to the majority of the plant species. It is found on contaminated and non-contaminated soils. It is considered as a model species to study adaptation to heavy metals. Multiple studies have focused on identifying the genetic basis of metal tolerance and hyperaccumulation in *N. caerulescens*, through QTL mapping and differential gene and protein expression analyses. Obviously, epigenetic studies are still scarce (Gulli, Marchi, Fragni, Buschini, & Visioli, 2018*) and are needed to add an additional layer to the comprehension of heavy metal tolerance in *N. caerulescens*. Our study aims at tracking epigenetics changes, particularly DNA methylation changes, across *N. caerulescens* populations from contaminated and non-contaminated sites, from two geographical locations: Luxembourg-Belgium and south-east of France, and under two experimental conditions: non- polluted and zinc-polluted conditions. We tracked methylation *via* Whole-genome Bisulfite Sequencing in the three different DNA contexts where it can occur: CpG, CHG and CHH, knowing that C-methylation in these contexts is regulated by independent pathways.

The genome-wide mean methylation rate was context-dependent: 53% for CpG sites, 19% for CHG sites and 11% for CHH sites. Zinc pollution applied in the greenhouse induced a genome-wide hypomethylation at the CHH sites relatively to the non-polluted condition, whereas the other contexts didn't show variation between the two conditions. Regarding latitude, we observed significantly lower CpG methylation and higher CHH methylation in northern populations. CpG methylation was significantly higher in populations from contaminated sites as opposed to CHG methylation which showed a reverse trend.

Gene body methylation is known to be mostly restricted to CpG sites, whereas non-CpG methylation also affects repeats and plays a key role in transposable elements regulation. In our case, the genome-wide response to instantaneous zinc pollution was CHH hypomethylation, suggesting an acute sensibility of transposable element regulation machineries in response to instantaneous environmental changes.

*Gulli, M., Marchi, L., Fragni, R., Buschini, A., & Visioli, G. (2018). Epigenetic modifications preserve the hyperaccumulator *Noccaea caerulescens* from Ni geno-toxicity. *Environmental and Molecular Mutagenesis*. doi:10.1002/em.22191

Massive premature cleavage and polyadenylation in a PRP31 mutant at 37°C

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In *Arabidopsis thaliana*, heat stress leads to a release of transcriptional repression of heterochromatin, independently of changes in patterns of common epigenetic marks. In a search for factors controlling this process, we have identified the *zen3* mutation affecting the *PRP31* gene, which encodes an essential splicing factor. In response to heat stress, *zen3* plants show compromised reactivation of heterochromatin transcription and mis-expression of thousands of genes. Transcriptome analysis reveals that this is associated with widespread early polyadenylation of mRNAs (PCPA for Premature Cleavage and PolyAdenylation) in *zen3*, suggesting an important role for PRP31 in protecting mRNAs from PCPA. Indeed, our survival tests attest of an essential function of PRP31. In order to assess potential role of PRP31 in protecting against PCPA, we used an artificial microRNAs implementation approach to discern a specific function in response to heat stress from a thermo-sensitive mutation. The detection of a GFP tag fused to wild type PRP31 or the mutant form by microscopy showed a clear altered nuclear distribution in *zen3* mutant.

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